RELATIONSHIP BETWEEN CONCENTRATION OF PLUM POX VIRUS AND CONTENT OF PIGMENTS IN VIRUS-INFECTED SYMPTOMATIC APRICOT LEAVES

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Summary. – Besides other factors, occurrence of plum pox virus (PPV)-caused spots and mosaic symptoms on leaves of stone fruits are known to influence important physiological functions including production of assimilates. Apricot (*Prunus armeniaca* L.) seedlings were used as a test material for typical manifestation of symptoms of the disease on the foliage. The relations of the abovementioned visual symptoms to the virus and pigments concentrations in leaves have so far not been known sufficiently. We detected PPV only in symptomatic leaf tissue of the infected apricot. In the green tissue of the same leaves, the double-antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) of the virus was negative. The content of pigments was changed by PPV infection. In the symptomatic leaf tissues, the content of chlorophylls "a" and "b" was lower, and the content of carotenoids was higher in comparison to the respective controls. We conclude that the PPV infection could cause the change in the content of particular leaf pigments leading to the decreased yield of photosynthesis which in turn could influence the sugar metabolism in the infected trees.

Key words: plum pox virus; apricot; symptomatic leaves; chlorophylls; carotenoids

Introduction

One of the characteristic symptoms of many plant virus diseases is mosaic spots on leaves. Plum pox (sharka) caused by PPV belongs to these diseases. The first symptoms of the disease appear on young leaves as irregularly distributed vein clearing. Later on, characteristic spots, ringspots and bands appear. Their size depends on the cultivar infected: the diameter varies from millimetres to a few centimetres. In the late phase of infection of some cultivars, the symptoms can appear also on fruits and woody parts (Németh, 1986). Besides other factors, the occurrence of PPV-caused spots and mosaic symptoms on leaves of stone fruits is known to influence important physiological functions including the production of assimilates (Visedo *et al.*, 1990, 1991).

Relations of the abovementioned visual symptoms to the virus concentration and the change of the content of leaf pigments have so far not been known sufficiently. The purpose of this work was to study the relationships between the concentration of PPV and the contents of chlorophylls and carotenoids in the virus-infected apricot leaves.

Materials and Methods

Host plant. The apricot (*Prunus armeniaca* L.) seedlings were used as a test material for typical manifestation of the disease symptoms on the foliage.

DAS-ELISA of PPV. Homogenates of symptomatic and green tissues of infected leaves (about 20 segments of 1 cm^2 size) were tested using a polyclonal antibody to PPV (Loewe Biochemica) according to the method of Clark and Adams (1977).

Content of pigments in the leaf lamina was assayed in the same segments which were used for the DAS-ELISA. Separation and assay of leaf pigments were carried out by thin layer chromatography according to Haspelová-Horvatovičová and Frič (1964), and Haspelová-Horvatovičová (1981). Briefly, the separation of pigments was carried out on silufol plates using benzene : isopropanol : water (100:10:0.25) as a separation liq-

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uid. Individual bands of separated pigments were quantitatively eluted into 2 ml of different elution liquids: chlorophylls "a" and "b" – acetone; lutein, violaxanthin and neoxanthin – 96% ethanol; β-carotin – n-hexane. The absorbance of eluted pigments was determined using a spectrophotometer Specord M 42. The content of pigments was calculated according to Goodwin (1955) using specific absorption coefficients.

Dry matter was determined by gravimetric method after desiccation at 105°C up to the constant weight. Experiments were done using three kinds of tissues: tissue of healthy leaves (control), tissue of symptomatic leaves, and green tissue of virus-infected leaves.

Results and Discussion

The obtained DAS-ELISA results (Table 1) indicated a positive reaction to PPV antibody of leaf tissues exhibiting yellow mosaic spots but a negative one of green parts of the virus-infected leaves and tissues of healthy control leaves. These results are in accord with the findings indicating that PPV is unevenly distributed in infected trees which can complicate sampling and detection of this virus (Albrechtová, 1986; Korschinek *et al.*, 1991; Polák, 1995; Knap *et al.*, 1995; Dicenta and Audergon, 1995). PPV could be detected only within the zone of chlorotic spots of leaves with symptoms (Fig. 1) in accord with the findings of Ranković and Vuksanović (1985).

In comparative experiments (Table 2), changes in the content of chlorophylls and carotenoids were evident in mosaic spot tissues but not in green parts of infected apricot leaves in comparison to healthy control leaves. A rapid decrease in the content of chlorophyll "a" and "b" was evident only in leaf tissues with mosaic spots. On the contrary, an increase in the content of carotenoids (especially neoxanthin and β-carotin) in mosaic spots and a decrease in the content of violaxanthin in green parts in comparison with healthy control was observed.

Pathological changes in the assimilation of pigments are in close relation to other physiological processes in the PPV-infected plants. Changes in the chlorophyll "a" : "b" ratio as well as in carotenoids, especially zeaxanthin-lutein and violaxanthin, are linked to changes in oxidation-reduction processes, mainly those involved in the photosynthesis. These changes cause the decrease of photosynthesis (Haspelová-Horvatovičová, 1981).

In PPV-infected susceptible cultivars of plum trees, the sugar content in fruits is reduced by about 0.14% – 2.11% and the content of acids is increased at the same time (Jordivič and Janda, 1963). Visedo *et al.* (1991) found that PPV-infected *Nicotiana clevelandii* leaves displaying symptoms of systemic infection showed moderate changes in the peroxidase isoenzyme profile that resembled the pattern of old leaves (senescence transformation).

It can be concluded from our results and the findings of other authors that the PPV infection could cause the changes in the content of particular leaf pigments leading to the decrease in yield of photosynthesis, which in turn influences the sugar metabolism in infected trees.

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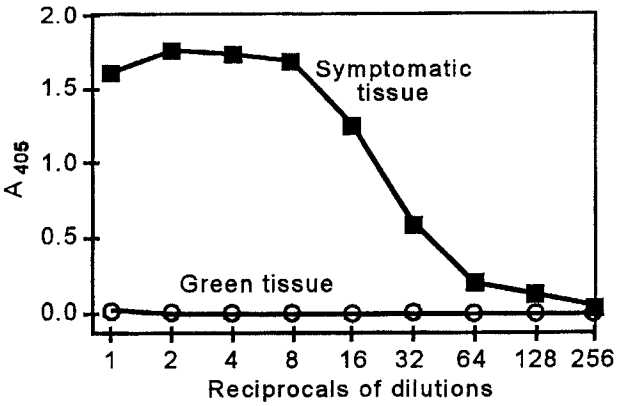

Fig. 1
DAS-ELISA of PPV
Calibration curves of homogenates of symptomatic and green tissues of PPV-infected apricot leaves.

Table 1. PPV concentration in homogenates of PPV-infected symptomatic leaves and green tissues of apricot

Apricot leaves	DAS-ELISA A_{405}	
Negative control (non-infected)	0.010	–
Positive control (infected)	1.695	–
Segments from diseased leaves with symptoms	1.532*	1.606*
Green tissue segments from diseased leaves	0 025*	0.017*

*Repeated experiments

Table 2. Influence of PPV infection on the content of leaf pigments in symptomatic apricot leaves

Leaf pigments	Pigment content (mg/g of dry matter)		
	Healthy leaves	PPV-infected leaves	
		Symptomatic tissue	Green tissue
Chlorofyll "a"	1.067	0.414	0.729
Chlorofyll "b"	0.474	0.168	0 337
β-carotin	0.063	0.130	0.083
Lutein	0.133	0.171	0.103
Neoxanthin	0.032	0.044	0.026
Violaxanthin	0.029	0.049	0.024

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